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New linkage data for group V markers in *N. crassa*

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Abstract

New linkage data for group V markers

for group V markers in *N. crassa*.

The genes rec-1 and rec-2 are located between ad-7 and asp on the right arm of linkage group V. It is likely that they are identical, since no recombinants have been detected in crosses having the form ret-1, rec-2 x + + (Catcheside, to be published). This would extend the known specificity of rec-1, which in previous tests was found to

affect allelic recombination at only his-1, of the ten loci examined (D. G. Catcheside and Austin 1969 Am. J. Botany 56: 685), to include nit-2, which is located on a different chromosome.

Since the assays for rec gene constitution are relatively laborious, mapping of one rec gene with respect to another has been expedited by bracketing the rec genes with closely linked markers, selecting for recombinants and scoring these for their constitution with respect to the unselected rec genes. It has consequently been of interest to locate markers close to rec-1. The data reported here were obtained in this search.

Materials and methods: Stocks of pab-2 (H193)A (FGSC#534), cot-2 (R1006t)A (FGSC#1513) and pyr-6 (DFC37)A (FGSC#2141) were crossed to a stock (t5242) having the genotype nit-2 (MN72), a, al-2 (15300); cot-1 (C102t); om-1 (47305); his-1 (K83), ad-7 (K77), asp (MN 137). Stocks of ro-4 (B38)A (FGSC#556) and ser-2 (65004)A (FGSC#2169) were crossed to t5246, a sibling of t5242 differing only in being nit-2⁺. The parents of cross 5327 were pab-2 (H193)A (FGSC#534) and on ro-4, asp isolate from cross 5258.

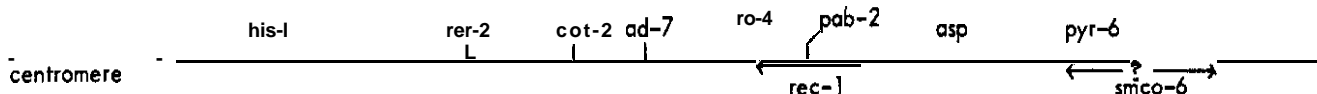
Crosses were made on modified Westergaard's medium (Catcheside 1970 Austr. J. Biol. Sci. 23: 855) containing the appropriate supplements (alanine 50, adenosine 20, asparagine 20, histidine 20, p-amino benzoate 1, uridine 10, serine 20, each expressed in mg/100 ml medium) and incubated at 25°C.

For cross 5258, ascospores were isolated onto slopes of fully supplemented medium (Vogel's salts, sucrose 2%, agar), heat shocked at 60°C for one hour and incubated at 25°C. ro-4 was scored by examining growth morphology, and other markers were determined by growth tests of conidial dabs on appropriately supplemented medium (Vogel's salts, sorbose 1%, sucrose 0.05%, agar.)

For all other crosses, selective plating was coupled with analysis of unselected markers. Heat shocked ascospores suspended in molten agar were diluted 1/5 and 1/25. The dilutions were plated on fully supplemented medium, the undiluted suspension and 1/5 dilution were plated on suitably supplemented selective plates (Vogel's salts, sorbose 1%, glucose 0.025%, fructose 0.025%, Difco agar) and incubated at 25°C. For cot-2, wild-type recombinants were selected by incubation at 34°C. The presence of cot-1 was taken into account in calculating recombination frequencies.

Unselected markers were scored in a random sample of recombinants. Small portions of colonies were isolated onto differential media (Vogel's salts, sorbose 1%, sucrose 0.05%, agar and supplements) and the plates were incubated at 25°C.

Results: The following map summarises the data presented in Table 1 and Table 2. Gene order is uncertain between intersecting lines. Arrowed lines indicate the approximate location of rec-1 and the most likely location of smco-6. Data are not reported for this latter gene since scoring of the smco phenotype was equivocal in 13 of the 70 random segregants examined.



No recombinants between ro-4 and pab-2 were observed in cross 5327. At the 95% confidence level, ro-4 is located within 0.23 map units distal and 2.8 units proximal to pab-2. This is in agreement with the findings of Perkins, Newmeyer, Taylor and Bennett (1969 Genetica 40: 247). If both sets of data are pooled, the results suggest that ro-4 is located within 0.19 units distal and 0.73 units proximal to pab-2.

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TABLE 1 Mapping data from selected recombinants

cross number	zygote genotype recombination % and deduced gene order				viable ascospores plated on each type of selective medium	number of selected progeny observed and unselected marker distributions				
						⊖ ad ⁺	⊖ asp ⁺	⊖ his ⁺	⊖ his ⁺ ad ⁺	⊖ ad ⁺ asp ⁺
5255	his-1	ad-7	⊖	asp	3080	117	100	221	113	6
	+	+	pab-2	+		his asp 13 his + 0 + asp 72 + + 5	ad 66 + 5	ad 41 + 51	asp 84 + 9	his 1 + 5
			7.6	6.5						
	—— 14.4 ——									
5261	his-1	⊖	ad-7	asp	2960	27	137	56	0	28
	+	cot-2	+	+		his asp 2 his + 18 + asp 0 + + 4	ad 69 + 23	ad 23 + 0		his 17 + 2
			7.6	3.6						
	—— 18.5 ——									
5269	his-1	ad-7	asp	⊖	2045	341	61	522	332	58
	+	+	+	pyr-6		his asp 5 his + 1 + asp 74 + + 16	ad 8 + 51	ad 36 + 60	asp 77 + 19	his 9 + 41
				6.0						
	—— 33.3 ——									
	—— ≈50 ——									
5272	his-1	⊖	ad-7	asp	4620	261	474	280	18	262
	+	ser-2	+	+		his asp 1 his + 90 + asp 0 + + 5	ad 42 + 54	ad 89 + 7	asp 1 + 15	his 90 + 6
			12.1	11.3						
	—— 20.5 ——									
5327	ro-4	⊖	asp		2675	-	104			-
	+	pab-2	+				ro-4 103 + 0			
			0	7.8						

TABLE 2 Mapping data from random spores

cross number	zygote genotype recombination % and deduced gene order				parental types	recombinants			total and germination %	
						singles †				doubles
						region 1	region 2	region 3		
	+	+	ro-4	+						
5258	his-1	ad-7	+	asp	25	7	2	0	0	74
					25	10	1	4	0	34%*
	23.0	4.1	5.4							

* less than 5% of ascospores from the cross failed to pigment.

† the upper number of each pair represents the class having the

+ allele of the leftmost marker for the interval.